9 MASS SPECTROMETRY

9.1 Objectives

9.1.1 Understand and explain the theory and application of mass spectrometry (MS).

9.1.2 Demonstrate a working knowledge of the design, operation and the components of a mass spectrophotometer.

9.1.3 Become proficient in the utilization of MS.

9.1.4 Generate and evaluate mass spectral information to confirm and quantitate the drugs being analyzed.

9.1.5 Interpret results by thoroughly explaining and comparing the mass spectra to libraries and databases.

9.1.6 Perform routine maintenance on the mass spectrophotometer.

9.2 Estimated Time: Three Months

9.3 Methods of Instruction

9.3.1 Lecture

9.3.1.1 Principles of mass spectrometry: ionization, source, detection

9.3.1.2 Modes of operation: total ion scan (TIC), selective ion monitoring (SIM)

9.3.1.3 MS components (sample inlets, ion sources, mass filters, detectors, vacuum systems)

9.3.1.4 Acquiring and evaluating mass spectra

9.3.1.5 Use of libraries and databases

9.3.1.6 Spectral interpretation

9.3.2 Literature Review

9.3.2.1 McLafferty, F. W., *Interpretation of Mass Spectra*, 3rd Ed. Chap 1.

9.3.2.2 Moffat, A. C., editor. *Clarke's Analysis of Drugs and Poisons*, 3rd Ed. The Pharmaceutical Press, London, 2004. pp 379-391.

9.3.2.3 Toxicology Technical Procedures Manual

9.3.2.4 Watson, J. T. *Introduction to Mass Spectrometry*. 3rd Ed. 1997. Lipincott-Raven.

9.3.2.5 Agilent Technologies GCMS Instrument Manuals

9.3.2.6 Mills, T and Robinson JC. *Instrumental Data for Drug Analysis*, 2nd Edition. Volumes 1-7, New York: Elsevier, 1987.

9.3.3 Demonstration

9.3.3.1 Operation of MS and analysis of specimens by MS will be observed from beginning to end and notes will be taken by the Trainee.

9.3.4 Laboratory Exercises

9.3.4.1 Inject the following drugs into the GC/MS individually and as a mixture: caffeine, codeine, monoacetyl morphine, diacetyl morphine, methadone, cocaine and methapyrilene. Determine the retention time of each drug. Identify each drug using the library. Determine the fragment ions generated for each drug. Compare the fragments with those in the library.

9.3.4.2 Analyze 10 unknown extracts to confirm the presence of alkaline extractable drugs by GCMS. Extracts from Section 7 (Extraction and Derivitization) may be used here.

9.3.4.3 Clean the ion source and evaluate the result.

9.3.4.4 Perform daily routine maintenance of the GC/MS to include but not limited to changing or adjusting the autotune, liner, septum, seals, gap column, transfer lines, gold seal, etc.

9.4 Evaluation

9.4.1 Written Exam

9.4.1.1 This will be administered as a “take home” exam.

9.4.2 Laboratory Competency Testing

9.4.2.1 Qualitative - a series of at least 10 unknown extracts will be presented to the Trainee for confirmation of the presence of alkaline extractable drugs.

9.4.2.2 Quantitative - At least 10 blood extracts will be presented to the Trainee for the quantitation of opiates, 10 for cocaine, and 10 for THC.

9.4.3 Courtroom Exercise

9.4.3.1 The Trainee must be capable of answering questions on this Module such as would be expected in a courtroom scenario.

9.5 Examination Questions

9.5.1 What is mass spectrometry?

9.5.2 Draw a schematic diagram for a GC/MS and describe the function of each component.

9.5.3 Diagram the EI source for the Agilent 5973.

9.5.3.1 Are the ions formed positive or negative?

9.5.3.2 Do they have an even or odd number of electrons?

9.5.3.3 What is the ionization efficiency of this technique?

9.5.4 What vacuum conditions are necessary in the ionization source and the analyzing regions of a MS and why?

9.5.4.1 Describe how a rough pump works.

9.5.4.2 Describe how a diffusion pump works.

9.5.4.3 Describe how a turbomolecular pump works.

9.5.5 Explain how chemical ionization is performed.

9.5.5.1 What are its advantages/disadvantages with respect to electron ionization?

9.5.5.2 What is the number of fragment ions produced by this method dependent on?

9.5.5.3 Do the ions formed by this process have an even or odd number of electrons?

9.5.6 Describe the difference between full mass scans and selective ion monitoring.

9.5.7 Describe how a quadrupole mass analyzer works.

9.5.8 Describe the importance of autotuning and explain the Autotune report.

9.5.9 Explain the function of the following: vacuum, ionization, filament, mass filter, and the electron multiplier.

9.5.10 Explain the following MS terms

9.5.10.1 mass to charge ratio

9.5.10.2 molecular ion

9.5.10.3 parent ion

9.5.10.4 base peak

9.5.10.5 total ion chromatogram

9.5.10.6 SIM

9.5.10.7 electron and chemical ionization

9.5.10.8 resolution

9.5.10.9 relative abundance

9.5.10.10 scan rate

9.5.10.11 spectral tilting

9.5.11 What is the effect of column bleed and/or septum bleed on GC/MS operation? What corrective action steps are normally taken?

9.5.12 How does the probability based matching library search work?

9.5.13 What reference spectra libraries are available in the toxicology section?

♦ **End**